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Prolonged antagonism of morphine-induced locomotor stimulation by κ opioid agonists: enhancement by prior morphine exposure

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Abstract

The selective κ agonists U50488 (10 mg/kg, i.p.) and spiradoline (1 mg/kg, i.p.) attenuated the locomotor activating effects of a morphine challenge (5 mg/kg, i.p.) administered 19 h later in rats. This antagonism of morphine by a κ agonist was reversed by the selective κ antagonist, norbinaltorphimine (10 mg/kg, s.c.). Furthermore, the κ opioid antagonism of morphine was enhanced by prior morphine exposure (2 doses of 30 mg/kg, i.p. administered once a day for 2 days). The present data suggest that κ - μ opioid interactions may occur over time periods that exceed the acute durations of drug actions.

Keywords: κ Opioid; μ Opioid; Morphine; U50488; Spiradoline; Locomotor activity

In recent years a substantial literature has shown that the effects of κ opioid agonists are often opposite to those of μ opioid agonists. For instance, κ agonists are considered to be dysphoric [24] and produce conditioned place aversions [20], while μ agonists produce feelings of euphoria and promote conditioned place preferences. Whereas κ agonists increase urination [17] and cause hypothermia [34], μ agonists induce anti-diuretic effects [6] and cause hyperthermia [35]. Furthermore, κ agonists have been shown to decrease dopamine release in the nucleus accumbens [8,9,32] and to decrease [18] locomotor activity, whereas μ agonists increase [8,9] dopamine release in the nucleus accumbens and increase locomotor activity [14].

Several studies have been performed to date which examined direct influences of κ agents on morphine-induced effects, or of μ agonist influences on subsequent κ opioid effects. For instance, it has been reported that a κ agonist can directly antagonize the effects of morphine in a Straub tail test [21], a morphine withdrawal study [11], a discriminative stimulus study [33] and in analgesia experiments [27]. Conversely, a μ agonist has been shown to antagonize the effects of a κ agonist in a shock titration procedure [5].

The few studies referred to above all examined acute interactions between μ and κ opioids. The present study sought to determine if a κ agonist could have a long term effect on subsequent morphine-induced locomotor activation. Previously, we reported that ibogaine, a naturally occurring alkaloid that has affinity for the κ receptor [7, 22], inhibited morphine-induced locomotor stimulation on the day after ibogaine administration [19,23]. Pretreating rats with morphine (1–4 daily injections) prior to ibogaine enhanced ibogaine antagonism of morphine [23]. Therefore, in the present study, the effect of prior μ agonist (morphine) exposure on subsequent κ agonist activity was also examined.

In the first study, saline pretreated (1 ml/kg, once a day for 2 days) rats were administered either a selective κ agonist U50488 (10 mg/kg, i.p.) or spiradoline (1 mg/kg, i.p.), or saline (1 ml/kg, i.p.) 19 h before being challenged with morphine (5 mg/kg, i.p.). Nineteen hours was selected because it corresponds with previous data in our laboratory using ibogaine. Locomotor activity was monitored for 3 h beginning immediately following the morphine challenge.

In the second experiment a separate group of rats received the selective κ antagonist norbinaltorphimine (norBNI, 10 mg/kg, s.c.) 1 h before U50488 (10 mg/kg, i.p.). Again, rats were challenged with morphine (5 mg/kg, i.p.) 19 h following the κ agonist or saline.

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In the third experiment rats were pretreated with either morphine (30 mg/kg, i.p.) or saline once a day for 2 days. Five hours after the second pretreatment injection, rats received either saline or U50488 (10 mg/kg, i.p.). Nineteen hours after the saline or κ agonist, rats were challenged with morphine (5 mg/kg, i.p.) and activity was monitored as described below.

Female Sprague–Dawley rats (250–275 g; Taconic Farms, Germantown, NY or Charles River, Kingston, NY, USA) were used for all studies. Rats were housed in groups of three or four, under a 12 h light cycle (lights on at 0700 h), in a room carefully controlled for temperature and humidity, and were provided food and water ad libitum. The use of animals in this study complies with the guidelines set by the Institutional Animal Care and Use Committee of Albany Medical College.

U50488 and spiradoline mesylate (RBI, Natick, MA, USA) were dissolved in sterile water at concentrations of 10 mg/ml and 1 mg/ml, respectively. Morphine sulfate (Mallinckrodt, St. Louis, MI, USA) was dissolved in saline at a concentration of 5 mg/ml. NorBNI (provided by Dr. S. Archer, Rensselaer Polytechnic Institute, Troy, NY, USA) was dissolved in 0.1 M NaH_2PO_4 at a concentration of 10 mg/ml.

Locomotor activity was monitored in cylindrical photocell activity cages (60 cm; three intersecting light beams). Data were recorded for the 3 h following the morphine challenge by a 386 computer with Med Associates software.

Statistical analyses were performed using analysis of variance (ANOVA) for treatment and time, with repeated measures over the times tested (Statistica). If ANOVA revealed specific effects then post hoc analysis was performed using the least significant difference (LSD) test.

In the first experiment ANOVA analysis revealed significant effects of drug treatment ($F_{(2,64)} = 8.3$; $P < 0.001$) and time ($F_{(2,128)} = 24.5$; $P < 0.000$). Subsequent LSD testing revealed that the κ opioid agonists, U50488 and spiradoline, significantly antagonized morphine-induced locomotor activity 19 h after their administration, as compared to rats receiving saline 19 h before the morphine challenge (Fig. 1).

In the second experiment ANOVA analysis revealed significant effects of treatment ($F_{(2,60)} = 6.3$; $P < 0.01$) and time ($F_{(2,120)} = 26.7$; $P < 0.000$). Subsequent post hoc analysis revealed that norBNI reversed the antagonism of morphine-induced locomotor activity by U50488 in the first hour only (Fig. 1, inset).

In the third experiment ANOVA revealed significant effects of treatment (U50488 or saline) ($F_{(1,71)} = 15.6$; $P < 0.001$) and time ($F_{(2,142)} = 35.1$; $P < 0.000$). LSD analysis showed that morphine pretreatment significantly enhanced U50488 antagonism (19 h later) of morphine-induced locomotor activity in the first and third hours following the morphine challenge, compared to rats which had been pretreated with saline before receiving

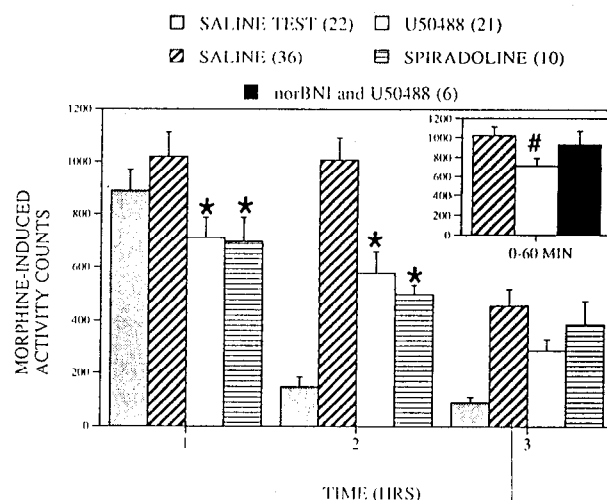


Fig. 1. The effects of the κ opioid agonists U50488 and spiradoline on morphine-induced locomotor activity 19 h after their administration. All rats were pretreated with saline (1 ml/kg, i.p. once a day for 2 days) and then received either saline (1 ml/kg, i.p.) or a κ agonist (10 mg/kg U50488, i.p. or 1 mg/kg, i.p. spiradoline) 19 h before receiving morphine (5 mg/kg, i.p.). A group of rats which received a saline test are shown to illustrate the extent of morphine-induced locomotor activation. The upper right hand inset graph illustrates the effects of a κ antagonist on κ agonist attenuation of morphine-induced locomotor stimulation. NorBNI (10 mg/kg, s.c.) was administered 1 h before U50488 (10 mg/kg, i.p.). # $P < 0.05$ compared to the U50488 group; * $P < 0.01$ compared to the saline group. Number of subjects are shown in parentheses and error bars indicate SEM.

U50488 (Fig. 2). Prior morphine exposure alone had no significant effect on a subsequent morphine challenge (compare Morphine Pre, Saline to Saline Pre, Saline group in Fig. 2).

The present study demonstrates that the selective κ agonists U50488 and spiradoline attenuate morphine-induced locomotor activity a day after their administration. This κ antagonism of morphine is reversed by pretreatment with the highly selective κ antagonist norBNI. Furthermore, prior morphine exposure in subchronic doses, enhances the κ antagonism of morphine-induced locomotor activity.

Antagonism of morphine-induced locomotor activity by κ agonists has been previously demonstrated [16] in an acute administration paradigm. In the present study, however, we report effects of κ agonists which occur a day after their administration and at much lower doses than previously demonstrated ([16]; 30 mg/kg U50488, i.p.). The authors are unaware of any pharmacokinetic studies at this time which indicate the half life of either κ agonist, and thus, cannot rule out the possibility that these drugs could persist in the brain at sufficient levels to produce the effects we see a day later. In support of this possibility, it has been previously reported that U50488 causes a reduction in food intake in rats 24 h after its administration [13]. If the drugs do not persist for long periods in the body, then perhaps they induce a long-lasting post-

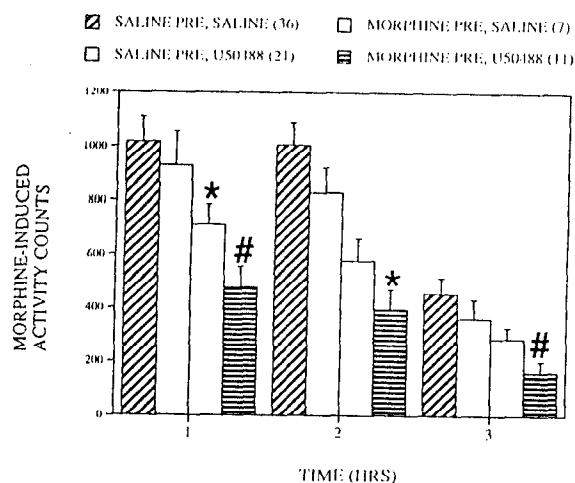


Fig. 2. The effects of prior morphine exposure on κ agonist antagonism of morphine-induced locomotor activity. Rats were pretreated with either saline or morphine (30 mg/kg, i.p. once a day for 2 days) and received either saline or U50488 (10 mg/kg, i.p.) 5 h after the last pretreatment injection. Nineteen hours after U50488 or saline, rats were challenged with morphine (5 mg/kg, i.p.) and activity was monitored. * $P < 0.05$ compared to Saline Pre, Saline group; # $P < 0.05$ compared to Saline Pre, U50488 group. Number of subjects are noted in parentheses and error bars denote SEM.

receptor change which interferes with morphine's ability to induce locomotor activity.

Locomotor activity is thought to reflect changes in dopamine in the nucleus accumbens (NAC) [15,26], and the present data might be accounted for in terms of μ and κ opioid influences on the mesolimbic dopamine system. Whereas μ agonists increase NAC dopamine [8,9], κ agonists acutely decrease dopamine in the same region [8,9,32]. Thus, U50488 and spiradoline may produce a long-lasting decrease in NAC dopamine which is sufficient to counteract the dopamine-increasing effects of morphine. An antagonism of morphine-induced dopamine release in the NAC by the κ agonists could explain the attenuation of morphine-induced locomotor activity seen in the present study.

The authors cannot rule out the possibility that the antagonism of morphine-induced locomotor activity by κ agonists could be attributed to a κ agonist induced tolerance at μ receptors. The literature regarding possible cross-tolerance between μ and κ opioids is inconclusive. There are several studies which report a 'small degree of cross-tolerance' [31] or complete cross-tolerance [10] between U50488 and morphine, while many others report no cross-tolerance between μ and κ opioid agonists [2,25,30]. However, this possibility seems unlikely because tolerance has never been demonstrated to occur to the locomotor stimulatory effects of morphine; rather, chronic morphine administration causes sensitization to the stimulatory effects of an acute morphine challenge [1,36]. Thus, a mechanism by which a single administration

of a κ agonist could induce tolerance to a later morphine challenge is elusive.

The current data also show that subchronic prior μ agonist (i.e. morphine) exposure can significantly enhance subsequent κ opioid antagonism of morphine-induced locomotor activity. The mechanism by which such an effect could occur is unknown. However, chronic morphine has been shown to up-regulate κ receptors [12], increase levels of dynorphin (1–13) in the brain [28] and up-regulate prodynorphin gene expression [29]; all of which could contribute to enhanced κ opioid activity. Similar to the results presented herein, it has been demonstrated that chronic morphine shifted a subsequent κ agonist dose-response curve to the left in analgesia studies [3,4] and that prior morphine exposure enhances the antagonism by ibogaine, which has affinity for the κ opioid receptor, of morphine-induced locomotor activity [22].

In summary, the present work demonstrates that complex interactions occur between μ and κ opioid agonists. Though pharmacokinetic data are not available, these interactions may occur over time periods that exceed the acute durations of drug actions.

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